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A Thesis
For the Degree of Master of Science

**Effects of Medium Chain Triglyceride with
Organic Acid on Growth Performance,
Blood Profiles, Nutrient Digestibility,
Intestinal Morphology, and Incidence of Diarrhea
in Weaning Pigs**

이유자돈 사료 내 중쇄지방산과 유기산의 첨가가
이유자돈의 성장성적, 혈액성상, 영양소 소화,
소장의 형태학적 변화 및 설사빈도에 미치는 영향

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이 논문을 농학석사 학위논문으로 제출함

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서울대학교 대학원 농생명공학부

고 태 옥

고태옥의 농학석사 학위논문을 인준함

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Summary

Weaning stress is one of the biggest stress during life of pigs. Due to weaning stress, situations such as low feed intake, body weight loss, and high incidence of diarrhea, occur to many piglets. It ultimately causes various diseases subsequently mortality rate increases which damages to many commercial farms. In order to solve this problem, AGP (antibiotic growth promoter) which is an antibiotic to be added to feed has been widely used. It can improve feed efficiency, promote animal growth, decrease incidence of disease, and enhance the quality of the animal products. Since antibiotic-resistant bacteria were found in humans as well as livestock, those were found to be propagated between humans and livestock the use of antibiotic as growth promoter has been forbidden in Sweden since 1986 for the first time in the world and prohibited in the EU in 2006 and in Korea in 2011, respectively. A number of alternatives have been proposed to overcome the increased mortality due to the ban on antibiotic use in feed. Numerous feed additives, such as enzymes, probiotics, prebiotics, plant extracts, and zinc, are typical examples. Among many alternatives, medium chain triglyceride (MCT) and organic acids (OA) are chosen because both MCT and OA have the common advantages of improving growth performance, having a strong antimicrobial effect and increasing nutrient digestibility. Therefore, it is needed to find optimal amount of MCT and OA in weaning pig to solve above problems. This experiment was conducted to evaluate the effects of medium chain triglyceride with organic acid on growth performance, blood profiles, nutrient digestibility, intestinal morphology, and incidence of diarrhea in

weaning pigs. A total of 120 crossbred ([Yorkshire x Landrace]) x Duroc) pigs (8.00 ± 0.870 kg BW) were assigned in five treatments considering sex and initial body weight in 3 replication with 8 pigs per pen in a randomized complete block (RCB) design. The first factor was two levels of MCT (0.1% or 0.2%), and the second factor was OA (0% or 0.1%). In the feeding trial, supplementing 0.1% MCT with 0.1% OA showed the greatest growth performance among treatments numerically. Also, supplementing 0.1% MCT with 0.1% OA especially improved ADG significantly during each phase (phase 1, $P=0.04$; phase 2, $P=0.01$; overall period, $P=0.01$) and increased G:F ratio significantly during phase 1 ($P=0.01$). In blood profiles, there were no significant differences in cortisol, IgG, TNF- α , IL-6, IL-10, and IL-1 β among treatments. Nutrient digestibility had no significant difference among treatments. The piglets fed 0.1% MCT treatment diet had greater villus height in duodenum and ileum, respectively during phase 1 (MCT, $P=0.04$; $P=0.03$). Also, 0.1% OA increased villus height and villus height to crypt depth ratio during phase 2 (OA, $P=0.01$; $P=0.04$). No significant difference was observed in the incidence of diarrhea among treatments. In conclusion, supplementing 0.1% MCT with 0.1% organic acid improved intestinal morphology of weaning pigs, thus it had positive influence on growth performance in pigs after weaning.

Key words: Medium chain triglyceride, Organic acid, Weaning pig,
Growth performance, Intestinal morphology

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III. Experiment

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List of Abbreviation

AA	Amino acids
ADG	Average daily gain
ADFI	Average daily feed intake
AOAC	Association of official analytical chemists
AGP	Antibiotic growth promoter
ATTD	Apparent total tract digestibility
BW	Body weight
CP	Crude protein
CRD	Completely randomized design
DM	Dry matter
EAAP	Experimental animal allotment program
EU	European union
FMD	Foot-and-mouth disease
FD ₄	Fluorescein isothiocyanate dextra
G:F	Gain : feed
GLM	General Linear Model
IgG	Immunoglobulin G
IL	Interleukin
LPS	Lipopolysaccharide
MCFA	Medium chain fatty acid
MCT	Medium chain triglyceride
ME	Metabolizable energy
NRC	National research council

OA	Organic acid
PUN	Plasma urea nitrogen
RCB	Randomized complete block
SAS	Statistical analysis system
SBM	Soybean meal
SO	Soybean oil
TER	Transepithelial electrical resistance
TNF- α	Tumor necrosis factor - alpha
VH : CD	villus height to crypt depth ratio

I. Introduction

Antibiotics have been widely used in feed for more than 60 years to increase growth performance and to prevent disease in livestock feeding environments (Smith, 1975; Vissek 1978; Kunnin et al, 1991; Anderson et al., 1999). However, the European Union (EU) has decided to prohibit antibiotics as growth promoter completely in 2006 since the bacteria resistant to antibiotics were found in human and livestock and antibiotic-resistant bacteria cross-infected between human and livestock (Casewell et al., 2003; Chen et al., 2005). In addition, Korea also has forbidden antibiotics as growth promoters from 2011. There are also a number of different reasons to minimize or completely eliminate the inclusion of in-feed antibiotic in livestock diets (Pluske, 2013). Consequences for the ban on antibiotics include less uniformity, a decrease in feed intake, body weight, feed conversion efficiency and an increase in incidence of diarrhea followed by high disease incidence and mortality rate of weaning pigs (Casewell et al., 2003; Pluske, 2013). To solve problems above mentioned previously during the post-weaning period, it is necessary to find an alternative of in-feed antibiotics since the prohibition. (Yen et al., 2015). There are many alternatives to AGPs such as probiotics, prebiotics, herbal extract, zinc oxide, organic acid (OA), and MCT (Dierick et al., 2002). Among many alternatives, MCT and OA are chosen as the alternatives because they have many common advantages like better growth performance, high nutrient digestibility and antibacterial effect in gastrointestinal tract of livestock. Therefore, it is expected to see synergy effects if MCT and OA are used together in the piglet diets.

Consequently, this study was conducted to evaluate the effects of MCT and OA supplementation on growth performance, blood profiles, nutrient digestibility, intestinal morphology and incidence of diarrhea in weaning pigs.

II. Review of Literature

1. Background

1.1 Problems of post-weaning stress in piglets

Weaning pigs get one of the most stressful weaning stress in their life by being separated from sow (Campbell et al., 2013). During the weaning, there are many stressors which include environmental, nutritional, social, and physiological stress that affect the life of pigs (Pluske et al., 1997). Moving to different and new physical environment (room, building, farm, etc.), eating different food source from sow milk to solid feed, co-mingling with pigs from other litters, experiencing social hierarchy stress, and increasing exposure to pathogens or environmental contamination are typical examples. As a consequence, weaning stress they get can cause intestinal and immune system disorders which result in reduced pig health, growth, and feed intake, particularly during the first week after weaning (Campbell et al., 2013).

Particularly, there are problems that the feed intake and growth are decreased and the inflammation occurs due to the stress of weaning. When the piglet is weaned, the piglet should now get used to new solid dry feed abruptly that is less digestible instead of adapting highly palatable liquid milk from its sow (Bruininx et al., 2001). Otherwise, this change causes piglet to eat less and become mal-nourished with poor growth rate. According to Le Dividich and Sève research (2000), metabolizable energy (ME) intake was decreased up to 60-70% of pre-weaning milk intake by the end of the first week post-weaning and it

needed 2 weeks post-weaning to go back to the pre-weaning ME intake level (Figure 1).

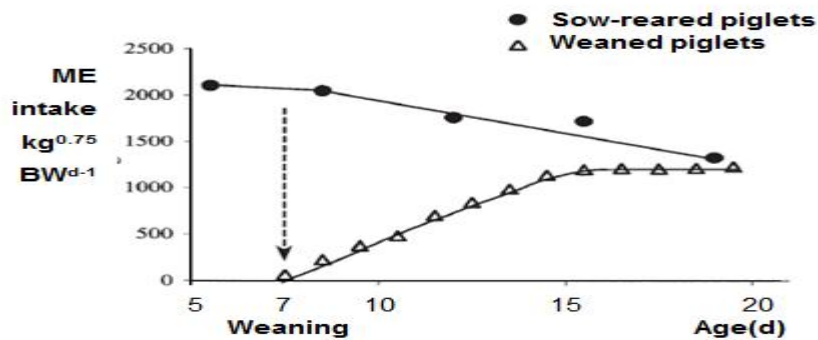


Figure 1. Daily amount metabolizable energy intake (ME) by suckling and weaning piglets (Marion et al., 2002)

In addition, low feed intake was associated with low growth performance. Le Dividich and Sève research (2000) reported that pigs lost about 20-30% of their relative body weight (BW) on the first day after weaning regardless of weaning age, and it took about four days to recover this loss in body weight. Tokach et al. (1992) reported that the total days to market (at approximately 110 kg BW) were highly affected by how much piglet grew in the first week after weaning. This was why it was important to pay attention to the feed intake and growth of piglets after weaning.

In addition to decreasing feed intake related to growth rate, weaning also caused inflammation which was harmful to intestinal barrier function (Spreeuwenberg et al., 2001). The epithelial layer of the intestinal lumen acted as a shield to protect pigs from various harmful microorganisms,

toxins or antigens. Figure 2 showed several types of proteins that connected cells and cells. However, as the intestinal barrier was weakened, harmful substances and bacteria could pass directly through the epithelial cells of the small intestine to the blood, resulting in inflammation, mal-absorption, swelling, dehydration, diarrhea and reduced growth (Redzic, 2011). In other words, permeability was increased due to destruction of intestinal barrier.

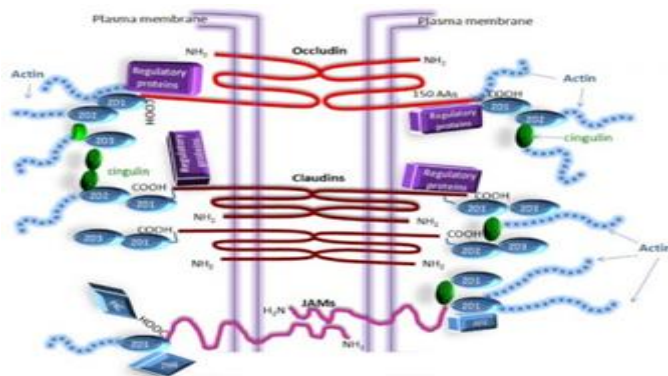


Figure 2. Schematic representation of the tight junction transmembrane proteins between two adjacent cells (Redzic, 2011)

Furthermore, Hu et al (2013) showed that the TER (transepithelial electrical resistance) of the jejunum decreased and FD4 (Fluorescein isothiocyanate dextra) in the mucosal and intestinal membrane increased from the weaning (Table 1). This meant that the villi and the crypts generally returned to the value of the weaning point at 14 days after the weaning, whereas the barrier recovery rate of the visceral mucosa was slow. In addition, the degree of the expression of the three mRNAs below indicated that the expression of the protein connecting the epithelial cells of the jejunum of the weaning piglets was lowered.

Table 1. Tight junction protein mRNA expressions and barrier function in jejunum after weaning

Item	Days post-weaning				SEM
	0	3	7	14	
TER¹-Ωcm²	65.8 ^c	51.4 ^b	48.7 ^b	53.5 ^b	2.95
FD₄ flux², ug cm⁻²h⁻¹	1.2 ^c	1.7 ^c	2.3 ^a	2.0 ^{ab}	0.12
Occudin	1.0 ^a	0.53 ^b	0.41 ^b	0.57 ^b	0.12
Claudin-1	1.0 ^a	0.64 ^b	0.53 ^b	0.61 ^b	0.09
Zonula occludin-1	1.0 ^a	0.62 ^b	0.72 ^b	0.79 ^{ab}	0.11

^{a-c} means within a row with different letters differ significantly (P<0.05).

¹ TER = Transepithelial electrical resistance

² FD₄ = Fluorescein isothiocyanate dextra (4kDa)

Other immune responses that occurred during the course of the weaning were changes in pro-inflammatory cytokine (Campbell, 2013). Pro-inflammatory cytokine were associated with permeability and transported of nutrients and thus affected intestinal integrity and epithelial function (McKay and Baird, 1999). The extent of cytokine mRNA expressions in the jejunal mucosa of piglets after weaning was presented in Table 2 (Hu et al., 2013). In this table, when pathogens or harmful substances entered into the body of weaning pig, several types of pro-inflammatory cytokine were secreted with the attack of macrophages which meant that the protective capacity of digestive organs became very weak. In case of piglet, the amount of feed intake after weaning was low and thus the capacity of the digestive organ was atrophied. As a result, the ability to defend harmful substances and pathogens could become very fragile if the digestive capacity was atrophied.

Table 2. Cytokine mRNA expressions in the jejunal mucosa of piglets after weaning¹

Item	Days post-weaning				SEM
	0	3	7	14	
TNF-α (Tumor necrosis factor- α)	1.00 ^c	4.48 ^a	2.97 ^{ab}	1.38 ^{bc}	0.56
IFN-γ (Interferon- γ)	1.00 ^b	3.04 ^a	1.94 ^{ab}	1.06 ^b	0.51
IL-6 (Interlukin-6)	1.00 ^b	3.85 ^a	3.08 ^a	1.31 ^b	0.45

^{a-c} means within a row with different letters differ significantly (P0.05).

¹ calculated relative to the value in samples from the pre-weaning (d 0 postweaning)

Johnson et al (1997) reported the results of TNF- α , IL-6, cortisol, and PUN after plasma injection of lipopolysaccharide (LPS) into the peritoneal cavity of the weaning pigs. In fact, the secretion of cytokine and cortisol could be confirmed to have increased. In addition, the large increase of PUN afterwards was to break down the body protein and use it for antibody production. Therefore, it could be seen from the table 2 and figure 3 that the protection capacity of the digestive organ of the weaned piglet was very shrinking.

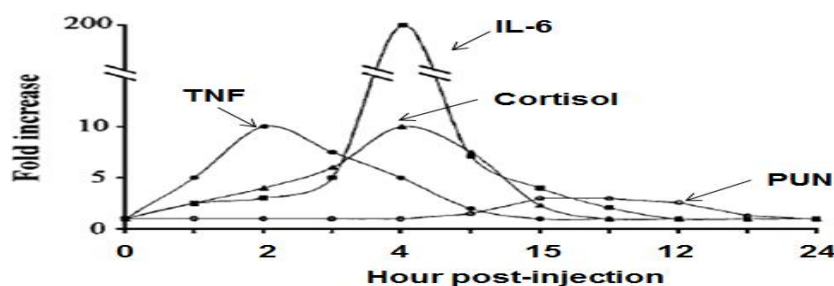


Figure 3. The plasma levels of TNF- α , IL-6, cortisol and urea nitrogen (PUN) following intraperitoneal injection of LPS (Johnson, 2011)

1.2 Structure and function of the small intestine

Weaning caused not only low feed intake but also morphologic and physiological changes in the structure and function (enzymatic activity and absorption or secretion) of the intestine (Miller et al., 1986; Cera et al., 1988; Dusford et al., 1989). These morphologic and physiological changes were highly associated with digestive and absorptive capacity of the small intestine which could have an influence on how much piglets can digest efficiently (Pluske, 1997). Many researchers have reported that these changes induced reduction in villous height (villous atrophy) and an increase in crypt depth (crypt hyperplasia) after weaning (Hampson, 1986; Pluske et al., 1996; Spreeuwenberg et al., 2001; Boudry et al., 2004; Lalles et al., 2004). Reduction in villus height could be explained by in terms of either an increased rate of cell loss or a reduced rate of cell renewal (Pluske, 1997). If villus atrophy occurred due to an former reason, then this would ultimately increase crypt depth with increased crypt-cell production (ex. microbial challenge, antigenic components of feedstuffs). On the other hand, if villus atrophy happened due to an latter reason, then this would be the result of reduced cell division in the crypts (ex. fasting).

Lalles et al (2004) demonstrated that not only villus height and crypt depth but also digestive enzymes and pancreatic enzymes (Table 3). They reported lactase and amino-peptidase activity was reduced from day 2 to 15 post-weaning, while maltase activity was decreased for 2 day post-weaning and then increased day 8 to 15 post-weaning. This may show whether intestine works well and fully grows or not related to the weaning diet. Regarding pancreatic secretions, trypsin and amylase activity

began to increase after day 8 post-weaning but lipase activity started to decrease at that time.

Table 3. Post-weaning changes in some architectural and functional parameters of the small intestine in young pigs weaned at 21 days of age

	Time post-weaning (days)		
	+2	+8	+15
Small intestine			
● tissue weight	- 18	+ 14	+ 39
● mucosa weight	- 20	+ 4	+ 46
Duodenum			
● villus height	- 40	- 37	- 23
● crypt depth	- 2	+ 41	+ 43
● digestive enzyme specific activities			
- lactase	- 19	- 71	- 80
- maltase	- 12	+ 2	+ 2
- amino-peptidase N	- 49	- 39	- 39
Pancreas			
● tissue weight	+ 2	+ 23	+ 57
● trypsin activity	+ 27	- 6	+ 65
● amylase activity	- 8	- 7	+ 23
● lipase activity	+ 35	- 59	- 61

1.3 Alternative additives for AGPs

Growth promotion and disease prevention have been effectively achieved by the inclusion of various antibiotics in the swine diets for more than 50 years (Thacker, 2013). However, the increased use of antibiotics in the swine diets has given a fear to consumers that they are concerned about resistant pathogenic bacterial strains (Budino et al., 2005; Vondruskova et al., 2010) and residual contamination of the food chain with antibiotics (Roselli et al., 2005; Van der Fels-Klerx et al., 2011). As a result, many countries have forbidden or are banning the use of

antibiotics as growth promoters in the diets (Chen et al., 2005). This movement has led to withdraw the antibiotic promoters from pig diets gradually and given consumers feel relaxed about meat safety. Due to the fact that antibiotics have been banned as growth promoters and more countries have participated to prohibit AGPs, an intensive research was conducted to develop antibiotic substitutes to maintain swine health and growth performance (Bomba et al., 2006; Castillo et al., 2008). Various natural materials which have been studied as efficient alternatives to antibiotic growth promoters include probiotics (Jacela et al., 2010; Simon, 2010; Cho et al., 2011), prebiotics (Jacela et al., 2010; Halas and Nochta, 2012), zinc oxide (Pettigrew, 2006; Jacela et al., 2010), plant extracts (Windisch et al., 2008; Liu et al., 2011), organic acids (Partanen and Mroz, 1999; Hansen et al., 2007), and MCTs (Zentek et al., 2014; Yen et al., 2015).

2. Properties of MCT

2.1 Chemical properties

Medium-chain Triglycerides (MCTs) are composed of a glycerol backbone and three saturated fatty acids which generally consist of 6-12 carbons. The fatty acids found in MCTs are called medium-chain fatty acids (MCFAs), such as caproic acid (C6:0, hexanoic acid), caprylic acid (C8:0, octanoic acid), capric acid (C10:0, decanoic acid) and lauric acid (C12:0, dodecanoic acid). MCFAs are fully saturated and unbranched monocarboxylic acids. They are also composed of C2-units (acetyl-CoA) and thereby have an even number of carbon atoms. MCTs have a smaller

molecular size, lower melting point, lower pKa value, lower energy density, and a comparatively high solubility in water compared to long-chain triglycerides due to a shorter hydrocarbon chain length. These distinct chemical properties differences between MCTs and long-chain triglycerides (LCTs) affect to how to digest, absorb, and metabolize. The standard chemical properties of caproic, caprylic, capric and lauric acid are shown in Table 4.

Table 4. Chemical properties of caproic, caprylic, capric and lauric acids

Fatty acid	Molecular weight (Da)	Melting point (°C)	pKa
Caproic acid (C6:0)	116.2	-3.4	4.88
Caprylic acid (C8:0)	144.2	16.7	4.89
Capric acid (C10:0)	172.3	31.9	4.89
Lauric acid (C12:0)	200.3	44.0	5.13

pKa, negative logarithm of the acid dissociation constant.
References: HSDB (2011), Hsiao and Siebert (1999).

2.2 Energy efficiency

One of the biggest characteristics of MCT is to deliver energy efficiently. MCT shows better energy efficiency than LCT. Figure 4 shows that MCTs with shorter chain lengths compared to LCT have the advantage of faster energy uptake and faster metabolism as energy resources. As a result of this accelerated metabolic conversion, instead of being stored as fat, the calories contained in the MCT are converted into fuels very efficiently and used immediately for organs and muscles. Also, figure 5 shows that MCTs pass through the two-layered mitochondrial membrane very quickly and do not require the presence of carnitine like

LCT. As a result, acetyl-coA is excessively produced along various metabolic pathways and then ketone body is produced in mitochondria and cytosol. Scientists reported that increased energy led to the rapid formation of ketone bodies in the consumption of MCTs. In other words, rapid energy production of MCTs means rapid production of the ketone body. Consequently, MCTs are a great choice for increasing energy efficiently.

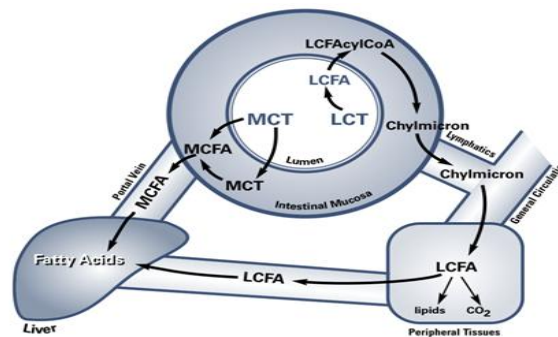


Figure 4. Digestion and transport of fats (Dean et al., 2013)

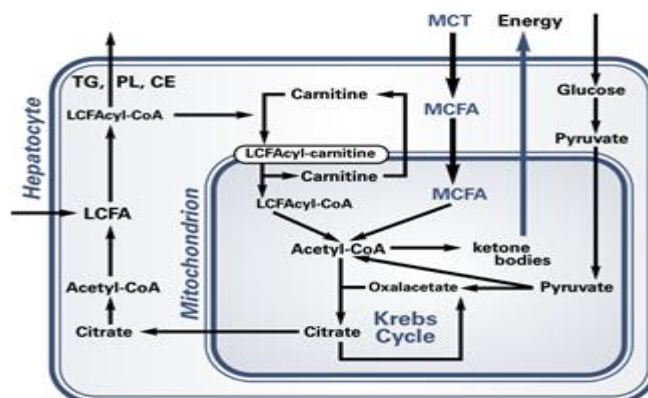


Figure 5. Metabolism of fatty acids in the liver and mitochondria (Dean et al., 2013)

2.3 Antibacterial effect

Medium-chain triglycerides have been shown to have antibacterial effects. Many research have found supplementing MCT could inhibit intestinal microbial count and thus cause piglets to improve growth performance (Lai et al., 2008; Yen et al., 2015). According to Yen et al (2015) experiment, piglets fed diets supplemented with 3% MCT had lower microbial counts compared to the control. All places including small intestine, cecum, colon and rectum showed lower microbial counts numerically compared to the control. Coliforms counts were especially decreased as MCT was supplemented in colon and rectum content ($P<0.05$). Furthermore, significant differences were observed in Dierick et al (2002) experiment. Four diets included : A (control), 2.5% soybean oil; B, 2.5% MCTAG2 oil; C, 2.5% MCTAG2 oil 11000 ppm lipase L5; D, 2.5% soybean oil 11.5% organic acids. The total anaerobic bacterial count and the counts of *Lactobacilli*, *Streptococci* and *E. coli* were given in the experiment. The results showed that there was a significant difference in the total bacterial load among treatments, both in the stomach and duodenal chyme ($P<0.05$). Similar result was true for *Lactobacilli* except for reaching significant difference. Also, Lim et al (2010) who conducted the experiment comparing the control with the treatment with MCT supplementation explained that the bacterial effect of MCT was highly related with prevention of diarrhea. In the experiment, no significant difference was observed in fecal coliform count among treatments. However, the farmers who used the MCT on piglet's diet had mentioned less diarrhea problem occurred in piglets fed MCT compared to piglets fed the control diet. A previous research conducted by Petschow et al (1998)

had proved that MCFA or their monoglycerides inactivated the pathogenic bacteria including *Pseudomonas spp.*, *Campylobacter spp.*, *Vibrio cholera*, *Salmonella typhimurium*, *Shigella sonnei*, *Hemophilus influenza*, *Listeria monocytogenes*, *Staphylococcus aureus*, *Streptococcus agalactiae*, *Helicobacter pylori* and enterotoxigenic *E.coli* and had antimicrobial effect on bacteria in gut to reduce the diarrhea problem in piglets. The bactericidal effect of MCT was mainly related to the hydrogen ion (H^+). The pKa value of MCFA was between 4.88 and 5.02 (Hsiao & Siebert, 1999; Palaprat et al., 2005), indicating that MCFA ionized and released H^+ and reduced intestinal pH (Rossi et al. 1998; Decuypere & Dierick 2003), inhibiting the growth of bacteria.

Consequently, MCT may reduce the number of harmful bacteria, increase the nutrients digestibility, strengthen immune responses, and finally improve the feed efficiency of piglets (Vervaeke et al. 1979; Dierick et al. 1986a,b; Jensen 1993; Chesson 1994; Anderson et al. 1999).

2.4 Properties of MCT and LCT

There are many differences between MCTs and LCTs. The differences come from the unique characteristics of MCTs. MCTs have several specific structural and physiological characteristics compared with LCTs (Takeuchi et al., 2006). Due to their chemical and physical properties, MCTs differ significantly from LCTs regarding digestion, absorption, and metabolism (Odle, 1997; Zentek et al., 2011). The main difference between MCTs and LCTs is summarized in Table 6 (Gent et al., 2016). LCTs need bile salts for emulsification which permit pancreatic lipases to be hydrolyzed efficiently and micelles containing LCFA and

monoglyceride digestion products to be formed due to lower water solubility of LCT. Also, LCTs depend on carnitine to enter mitochondria since the chain length of LCT is longer than MCT. However, due to the relatively higher water solubility, MCT is hydrolyzed rapidly compared to LCT without the necessity for emulsification with bile and lipase. In addition, MCTs are independent on carnitine to enter mitochondria because MCTs can pass mitochondria membrane very rapidly. Likewise, absorption of MCT is faster and oxidation of MCT is higher than LCT. In other words, after being absorbed, MCFA can be transported via a portal vein directly to the liver, with no re-esterification or chylomicron formation and preferential oxidation in the mitochondria (Bremer, 1980). It can thus be used more efficiently as an energy source compared to long-chain fatty acids (Odle, 1997).

Table 5. The comparison of main properties difference of MCTs and LCTs

Properties	LCTs	MCTs
Water solubility	Relatively insolubility	High solubility
Digestion	Necessity for emulsion with bile and require lipase	Without the necessity for emulsion with bile and lipase
Absorption	Reach the bloodstream via the lymphatic system	Absorbed predominantly via the portal vein into the liver
Metabolism	Require binding to fatty acid-binding protein, fatty acid transport proteins or fatty acid translocase (FAT); oxidation mainly occur in muscle tissue and most be stored in adipose tissue	Do not require binding to fatty acid-binding protein, fatty acid transport proteins or fatty acid translocase (FAT); oxidation mainly occur in liver and less be stored in adipose tissue
Enter mitochondria	Depend on carnitine	Independent on carnitine
Oxidation rate	Slow	Fast

3. MCT supports the integrity of the intestine

3.1. Effects of MCT on intestinal epithelial mucosal structure

Many researchers reported that growth performance of piglets was improved by due to the better digestibility of fat and protein in the diet including MCT. This turned out to be the result of improving intestinal epithelial mucosal structure (Hanczakowska, 2017). The mucosal epithelium of the small intestine played important roles for nutrient degradation and absorption (cera et al., 1988). Functional surface area of the mucosa was increased by villi and crypts. Villi have finger-like protrusions in the epithelial inner layer that help to increase the surface

area for digestion and absorption processes (Fuller, 2004), while crypts are the tubular glands located on the mucosal surface of the small intestine that enter the lumen at the bottom of the villi and contain epithelial stem cells required for regeneration of epithelial cells (Llyod and Gabe 2008). In addition, the bottom of the crypts is constantly fragmented to maintain the structure of the villi (Loh et al., 2013). Since the epithelial cells near the villus tip are the best developed, maturity is the highest and digestive and absorptive capacity are also the greatest (Tang et al., 1999). Consequently, the growth performance of piglet is highly related with villus height (Zijlstra et al., 1996).

Their relationship was found in many previous experiments (Dierick et al., 2003; Mekbugwan and Yamauchi, 2004; Hanczakowska et al., 2011). In Dierick's experiment, piglets fed diets with *Cuphea* seeds with a lipase supplement as a source of MCFA grew well as well as significantly greater villus height and lesser crypt depth were shown. Similar results were found in other research done by Mekbugwan and Yamauchi (2004). They found a highly positive relation between an increased villus height and improved growth performance of piglets. Hanczakowska et al. (2011) also compared both caprylic and capric acids fed separately or together with the control. Treatments including caprylic acid or capric acid or both showed significantly higher body weight and ADG and resulted in greater villus height but significant difference was only observed in the case of capric acid compared to the control group.

3.2. Effects of MCT as an immuno-modulator

MCT have been suggested to improve gut health under inflammatory

conditions. Stress of weaning pigs causes the integrity of the epithelial cells in the small intestine to be loosened. Then, this makes it easy for undigested nutrient, harmful bacteria and toxins to penetrate into the blood and lymph. At this time, macrophages, which are known as phagocytes ingest above materials and secrete substances called cytokine. The cytokine contact the secondary immune cells such as T and B cells to make antibodies to neutralize harmful bacteria or toxins. The reason for the low secretion of cytokine in piglets fed MCT is that MCT sterilizes noxious bacteria. Then, macrophages may secrete less cytokine and the villi of the small intestine grow well and long when harmful bacteria are killed. This process is called an immune-modulator. In addition, an increase in cytokine when a substance is added can be said to be an immune-modulator.

4. MCT on piglet performance

4.1 Effects of MCT on the growth performance of piglet

There are many research about the effects of MCT on the growth performance of piglets. Many research reported supplementing MCT on weaning pig diets would result in improving growth performance of weaning pig especially during phase 1 (Dierick et al., 2002; Hong et al., 2012; Li et al., 2015; Yen et al., 2015).

In the Dierick et al. (2002) study, the growth rates of the two treatments with MCT supplementation were significantly different with other treatments. This experiment consisted of four different diets : A (control), 2.5% soybean oil; B, 2.5% MCT; C, 2.5% MCT + 1000 ppm

lipase; D, 2.5% soybean oil + 1.5% organic acids. As a result, growth rates of treatments B and C with MCT supplementation were significantly higher than those of other groups ($p < 0.05$). Also, no significant difference was observed in feed intake and FCR but treatments B and C with MCT supplementation showed numerically higher feed intake and better FCR than the other two treatments. The other experiment conducted by Hong et al. (2012) showed similar results with Dierick's study. Researchers conducted two experiments with different 21 d and 28 d of age weaning pigs. In both of experiment 1 and experiment 2, the same dietary treatments were supplemented as follows : 1) negative control (NC), 2) positive control (PC), NC + antibiotics (40 mg/kg Tiamulin, 110mg/kg Tylosin, and 10 mg/kg Enramycin, 3) MCT3, NC + 0.32% (phase 1, 2 and 3) MCT and 4) MCT5, NC + 0.55% (phase 1), 0.32% (phase 2 and 3) MCT. Consequently, the ADG was higher in MCT5 treatment than NC treatment during phase 1 ($p < 0.05$) and was highest in MCT5 among other treatments as well in experiment 1. Also, MCT3 and MCT5 treatments showed higher ADG significantly in phase 1 than NC and PC treatments in experiment 2 ($p < 0.05$). According to Li et al (2015) experiment, weaning pigs fed the diets containing MCTs all grew up better than piglets in the control. Dietary treatments included : control diet [containing 3.5% soybean oil (SO)], MCT1 diet (containing 0.7% MCTs and 2.8% SO), MCT2 diet (containing 1.4% MCTs and 2.1% SO) and MCT3 diet (containing 2.1% MCTs and 1.4% SO). The purpose of this experiment was to find out the optimal amount of MCT in weaning pig's diet. As a result, treatments with MCT supplementation all improved ADG (linear, $p = 0.011$; quarartic, $p = 0.041$) and feed efficiency (linear,

p<0.01) compared with the control. In all experiments above, significant difference was observed in growth performance among treatments and treatments including MCT showed better growth performance compared to the control during phase 1.

The reason for these results was due to the special characteristics of the MCT. Unique physiological and biological properties of MCT resulted in improved growth performance of weaning pigs (Li et al., 2015). Piglets could digest and absorb MCT easily which could be effectively used as supplying energy by oxidation (Ode et al., 1991). This happened because MCTs could go through the double mitochondrial membrane very rapidly without carnitine and that was why MCT could be used as energy source immediately which could be supplemented to the diet of weaning pigs. Furthermore, MCT had anti-bactericidal activity which could reduce bad pathogens to utilize nutrients effectively. Harmful bacteria like *E.coli* required nutrients for their own growth and use but MCT could use their energy more on improving growth performance and digestibility by inhibiting of the proliferation of pathogenic bacteria.

4.2 Effect of MCT levels on apparent total tract digestibility of nutrients of weaning pigs

In many previous studies related to MCT experiments, researchers chose apparent total tract digestibility as a measurement to see whether MCT really could help piglets digest and absorb nutrients easily or not. Many research reported supplementing MCT on weaning pig diets would result in improving nutrient digestibility of weaning pig (Han et al., 2010; Hanczakowska et al., 2011; Hong et al., 2012; Li et al.,

2015).

According to Han et al (2010) experiment, there was significant difference in the MCT treatment. Four treatments consisted of a basal diet supplemented with antibiotics (33 mg per kg tiamulin and 44 mg per kg lincomycin), ZnO (1500 or 2500 mg per kg), or 0.1% E-MCFAs. As a result, weaning pigs fed the diet supplemented with MCT had the highest nutrient digestibility on dry matter, crude protein, calcium, phosphorus, and energy compared to other treatments ($p<0.01$). In Hanczakowska et al (2011) experiment, treatments including MCFAs showed better apparent digestibility compared to the control. The piglets were fed a basal diet (control), basal diet + 2 g of C₈ (caprylic acid), basal diet + 2 g of C₁₀ (capric acid) and 1 g of C₈ + 1 g of C₁₀, respectively. Consequently, the treatments including C₈, C₁₀, and C₈+C₁₀ which were one of fatty acids found in MCT showed significant differences on crude protein ($p<0.01$) and crude fiber ($p<0.05$) compared to the control. Li et al (2015) also conducted an apparent total tract digestibility in different levels of MCT supplementation experiment. In this experiment, there were significant differences in dry matter (linear, $p=0.032$) and ether extract (linear, $p=0.004$) during day 12 to 14 post-weaning. Furthermore, an increased ATTD of dry matter was found in the MCT2 treatment ($P<0.05$) and an improved ATTD of ether extract was observed in both MCT2 and MCT3 groups during day 12-14 post-weaning compared to the control ($P<0.05$).

As a result of ATTD in MCT experiment, MCT affected nutrient digestibility positively. This result was observed because MCT may

alter morphology of the small intestine, reduce bacteria, and ameliorate fat mal-absorption. Firstly, MCT may influence nutrient digestibility by changing villus height and crypt depth which were intestinal morphology of piglets (Cera et al., 1988; Li et al., 1990; Hanczakowska et al., 2011). Villi which were indicator of the functional capacity of enterocytes has been higher when MCT were supplemented. This meant that villi could increase the surface area for greater digestive and absorptive capacity (Hampson, 1986; Fuller, 2004). Secondly, one of characteristics of MCT that could reduce bacteria because of antimicrobial effects may influence on improved nutrient digestibility (Hong et al., 2012). Lastly, MCTs could improve the fat mal-absorption in reduced absorptive surfaces or atrophied intestinal villi.

4.3 Effects of MCT on the blood profiles of piglet

There are many different measurements related to blood profiles in the MCT experiment. BUN (blood urea nitrogen), albumin, cortisol, many interleukin, IgG, WBC, RBC etc were measured in various MCT experiments. Based on various research, positive results of blood profiles were presented by MCT factor.

In Han et al (2010) experiment, there were significantly differences in zinc, glutamic-oxaloacetic transferase and glutamic-pyruvic transferase ($p < 0.01$). The purpose of this experiment was finding the best alternatives of AGP which were forbidden in many countries. There were four options for alternatives such as antibiotics, ZnO 1500

mg/kg, ZnO 2500 mg/kg, and MCT. As a result, serum zinc, glutamic-oxaloacetic transferase and glutamic-pyruvic transferase concentrations were lower for piglets fed diets supplemented with MCT. Using the pharmacological level of Zn from ZnO has been suggested to be effective in controlling *E. coli* in weaning pig (Holm, 1988; Poulsen, 1989). However, many countries tried to reduce the dietary supply of Zn to consider environmental problems in some areas of intensive pig farming because highly inclusion of Zn in pig diets consequently caused toxicity to plants and microorganisms (Coppenet et al., 1993; Jondreville et al., 2003). Also, higher levels of glutamic-oxaloacetic transferase and glutamic-pyruvic transferase concentrations would be related with liver damage, so significantly the lowest value of those enzymes in MCT treatment would mean good for liver health. The other experiment conducted by Li et al (2015) showed significantly differences in plasma urea nitrogen and plasma total protein during the first 2 weeks post-weaning. Plasma TP is routinely used as an indicator for animal health, and plasma UN is the main end product of protein metabolism. The changes in these parameters may reflect the status of protein metabolism and utilization in animals (Eggum, 1970). In this experiment, pigs fed the MCTs-rich diets on the first 2 weeks post-weaning showed increased concentration of plasma total protein and decreased of concentration of plasma urea nitrogen significantly. These results showed increased concentration of plasma TP due to higher ATTD of CP during phase 1. Also, lower plasma UN concentration showed a better nitrogen balance after MCTs treatment.

As a result, many measurements were shown in many experiments with good results when MCTs were supplemented. Many measurements meant that MCTs were effective in many blood profiles and also needed to be conducted more research.

5. Combinations of MCT and organic acid

MCT and organic acid are known to have a significant effect when used individually, and there are many common positive points such as improving growth performance, reducing harmful bacteria, and increasing nutrient digestibility. However, there are not many research for supplementing MCT with organic acid in the weaning pig's diets. It can highly be expected that synergy effect might appear by using MCT with organic acid in diets for piglets especially improvement of digestive processes and intestinal micro-organisms.

In Kuang et al (2015) experiment, significant difference was found in body weight ($p<0.01$), average daily feed intake ($p<0.01$), apparent ileal digestibility of a majority of dietary amino acids ($p<0.01$), ileal ($p<0.01$) and rectal *Lactobacillus* ($p<0.05$). Treatments were 1) CON : basal diet with supplemental antibiotics (containing pure colistin sulfate and enramycin, respectively, at 0.02 g/kg diet) and ZnO (2.5 g/kg diet) 2) SF3 (containing calcium formate, calcium lactate, citric acid and MCFA at 340, 160, 70 and 130 g/kg, respectively). The piglets fed MCT with OA had higher ADFI ($P<0.01$) during the experimental weeks 1 and 2, BW ($p<0.01$) during from week 1 to 3, ADG ($p<0.01$) during week 1 and G:F ratio ($p<0.01$) during week 1 than

the piglets fed control diet. All measurements about growth performance improved when mixture of MCT and OA were fed. This could be the results because of few reasons. Firstly, MCT could be effective in supplying energy for growth performance. Secondly, formic acid and potassium diformate could increase feed intake and growth rate (Lallès et al., 2009; Partanen, 2001). Lastly, organic acid could prevent weaning induced stress and give great potential for growth rate if used together with MCT. Furthermore, SF3 treatment showed higher AID of a majority of amino acids (AA) including methionine, lysine, threonine, valine, phenylalanine, leucine, isoleucine, histidine, aspartic acid, glutamic acid, serine and tyrosine than the control ($p<0.01$). This happened because AA transporters were helpful in AA absorption and metabolism (Palacín et al., 1998). Also, piglets fed SF3 diets had higher ileal ($p<0.05$) and rectal *Lactobacillus* ($P<0.10$). This would suggest that microbiota diversity was affected positively by MCT and OA supplementation. *Lactobacillus* also had antibacterial effect against pathogenic strains such as *E.coli* and *Salmonella* (Tamura, 1983; Muralidhara et al., 1977). Lower plasma tumor necrosis factor alpha and higher plasma IgG concentrations were also found in SF3 fed piglets than the piglets fed control. Plasma tumor necrosis factor alpha was decreased in this experiment and this would suggest piglets might grow more and consume more feed intake immunologically. Furthermore, the increased plasma IgG concentrations meant that weaning pigs were stronger against harmful bacteria and immunity were also increased by dietary combinations of MCT and OA consumption. On the basis of these results, the increased *Lactobacillus*

could be explained with decreasing pro-inflammatory cytokine such as tumor necrosis factor alpha and IL-6 in the colon (Chen et al., 2005).

Not many research was conducted so far about supplementing MCT with organic acid on weaning pig's diet. However, positive possibility for this study was found and more constant research is needed.

III. Effect of Medium Chain Triglyceride with Organic Acid on Growth Performance, Blood Profiles, Nutrient Digestibility, Intestinal Morphology, and Incidence of Diarrhea in Weaning Pigs

Abstract: This experiment was conducted to evaluate the effect of medium chain triglyceride with organic acid on growth performance, blood profiles, nutrient digestibility, intestinal morphology, and incidence of diarrhea in weaning pigs. A total of 120 weaning ([Yorkshire x Landrace] x Duroc) pigs (8.00 ± 0.870 kg BW) were assigned in five treatments considering sex and initial body weight in 3 replication with 8 pigs per pen in randomized complete block (RCB) design. The first factor was two levels of MCT 0.1% or MCT 0.2%, and second factor was OA 0% or OA 0.1%. In the feeding trial, supplementing 0.1% MCT with 0.1% OA showed the greatest growth performance among treatments numerically. Also, supplementing MCT especially improved ADG significantly during each phase (phase 1, $P=0.04$; phase 2, $P=0.01$; overall period, $P=0.01$) and increased G:F ratio significantly during phase 1 ($P=0.01$). In blood profiles, there was no significant difference in cortisol, IgG, TNF- α , IL-6, IL-10, and IL-1 β among treatments. Nutrient digestibility had no significant difference among treatments. The piglets fed with 0.1% MCT had greater villus height in duodenum and ileum, respectively during phase 1 (MCT,

P=0.04; P=0.03). Also, 0.1% OA increased villus height and villus height to crypt depth ratio during phase 2 (OA, P=0.01; P=0.04). No significant difference was observed in the incidence of diarrhea among treatments. In conclusion, supplementing 0.1% MCT with 0.1% organic acid improved intestinal morphology, thus it had positive influence on growth performance in weaning pigs.

Keywords : Medium chain triglyceride, Organic acid, Weaning pig,
Growth performance, Intestinal morphology

Introduction

Weaning pigs experience one of the most stressful events when they are weaned from the sow (Campbell et al., 2013). In order to solve this problem, AGP has been widely used but it was forbidden because of many side effects (Marshall and Levy, 2011; Stanton, 2013). A number of alternatives have been proposed to overcome the increased mortality due to the ban on antibiotic use in feed (Seal et al., 2013). Among many alternatives, MCT and organic acids seem very effective to solve above problems.

MCT has particular nutritional and metabolic effects including fast digestion, passive absorption, mandatory oxidation, and is therefore particularly useful for nutritional ingestion of piglets (Odle, 1997). It has been reported in many previous studies that MCT has a positive effect in improving growth performance of piglets especially during phase 1 (Rodas and Maxwell, 1992; Dierick et al., 2003; Hong et al., 2012, Li et al., 2015). In addition, MCT has antimicrobial effects on *Gram-positive cocci* (Bergsson et al., 2001) and *Escherichia coli* (Skrivanová et al., 2006; 2009) and also protective effects on intestine microstructure (Dierick et al., 2003).

Organic acid and their salts especially formic acid and potassium formate have the advantage of improving feed intake and growth rate of piglets (Lawlor et al., 2006). They are used to preserve feed and affect the gastrointestinal environment and the digestive process (Kirchgessner and Roth, 1988; Partanen and Mroz, 1999). In addition, organic acids and their

salts also have the effect of preventing diarrhea in weaning pigs (Franco et al., 2005; Halas et al., 2010). Moreover, there is an effect of intracellular acidification, and another possible action mechanism is to inhibit bacteria (Partanen and Mroz, 1999).

Although both MCT and organic acid have many advantages, there are not many studies on supplementing both MCT and organic acid at the same time. Synergy effects are expected if MCT and organic acid are used together in the piglet diets. Therefore, this study was conducted to evaluate the effects of MCT with OA on growth performance, blood profiles, nutrient digestibility, intestinal morphology and incidence of diarrhea in weaning pigs.

Materials and Methods

Experimental animals and management

A total of 120 weaning ([Yorkshire x Landrace]) x Duroc) pigs (8.00 ± 0.870 kg BW) were allotted to one of five treatments considering sex and initial body weight in 3 replication with 8 pigs per pen in a randomized complete block design. Pigs were randomly allotted to their respective treatments by EAAP (experimental animal allotment program; Kim and Lindemann, 2007). Pen was fully-concrete floor facility (1.54×1.96 m) in experimental period and equipped with feeder, water nipple, and environmentally controlled facility in Seoul National University Farm. The experimental period was 5 week. Experimental period consisted of 2 phases, phase 1 was 0-2 week and phase 2 was 3-5 week.

Experimental design and diet

The first factor was two levels of MCTs (MCT 0.1% or MCT 0.2%), and second factor was with or without OA (OA 0% or OA 0.1%). Dietary treatments included : 1) CON (Corn-SBM based diet), 2) LM (Corn-SBM based diet + MCT 0.1%), 3) LMO (Corn-SBM based diet + MCT 0.1% + OA 0.1%), 4) HM (Corn-SBM based diet + MCT 0.2%), 5) HMO (Corn-SBM based diet + MCT 0.2% + OA 0.1%). Experimental diets were formulated for 2 phases, including weaning phase 1 and weaning phase 2. All nutrients of experimental diets except CP and ME were met or exceeded the nutrient requirement of NRC (1998). ME was determined to meet NRC 2012 and CP was set by multiplying the total nitrogen of NRC 2012 by 6.25. Formula and chemical composition of

experimental diet were presented in Tables 1 and 2.

Growth performance

Body weight, and feed intake were collected at the end of each phase in order to calculate average daily gain (ADG), average daily feed intake (ADFI) and gain to feed ratio (G:F ratio). In addition, feeding to all piglets were recorded each day, and waste feed in feeder was recorded on the end of each phase.

Blood sampling and analyses

Blood samples were taken from the jugular vein of five pigs near average body weight in each treatment after 3 hours fasting for measuring cortisol, IgG, TNF- α , IL-1 β , IL-6, IL-10 when the body weights were recorded. Collected blood samples were centrifuged for 15 min at 3,000 rpm on 4 °C (Eppendorf centrifuge 5810R, Germany). The sera were carefully transferred to 1.5 ml plastic tubes (Serum tubes, BD vacutainer® SST™ II advance, UK) and stored at -20 °C until analysis. To investigate the degree of stress, cortisol concentration was analyzed using an ELISA assay (Microplate Reader, VERSA Max, Swine Cortisol ELISA Kit, USA) and IgG concentration was measured by an ELISA assay (Microplate Reader, VERSA Max, Pig IgG ELISA Kit, USA) to determine immune status. To assess the degree of inflammatory cytokine changes, TNF- α concentration was measured by an ELISA assay (Microplate Reader, VERSA Max, Quantikine Porcine TNF-a / TNFSF2 Immunoassay, USA), IL-1 β concentration was measured by an ELISA assay (Microplate Reader, VERSA Max, Quantikine Porcine IL-1 β / IL-1F2 Immunoassay,

USA), and IL-6 concentration was measured using an ELISA assay (Microplate Reader, VERSA Max, Porcine IL-6 Immunoassay, USA). To investigate the degree of anti-inflammatory cytokine changes, IL-10 concentration was measured by an ELISA assay (Microplate Reader, VERSA Max, Quantikine Porcine IL-10 Immunoassay, USA).

Nutrient Digestibility

A total of 15 crossbred barrows, averaging 12.48 ± 0.37 kg body weight were allotted to individual metabolic crate (40 x 80 x 90 cm) in completely randomized design (CRD) with 3 replicates to evaluate the nutrient digestibility and nitrogen retention. Total collection method was used for the apparent total tract digestibility (ATTD; McCarthy et al., 1973). After 5 days adaptation period, 5` days of collection period was followed. To determine the first and last day of collection days, 8 g of iron oxide and chromium oxide were added in the first and last experimental diet as selection markers. During the experimental period, all pigs were fed 256 g of experimental diets twice a day which was three times the maintenance energy at 7:00 and 19:00, and water was provided *ad libitum* (Kim et al., 2012). Feces were collected by using total collection method and urine was collected daily in a plastic container. Feed intake, feces and urine were recorded everyday. Collected feces and urine samples were stored -20°C until analysis. Collected excreta were pooled and dried in an air-forced drying oven at 60°C for 72 hours, and then grounded into 1mm particles in a Wiley mill for chemical analysis. In case of urine, sulfuric acid solution, which was a role of collecting ammonia by chemical reaction, was titrated from 99% sulfuric acid

solution to 10% sulfuric acid solution and was put into 50 ml plastic case. Glass wool was put into the funnel that was stuck on the plastic case to make the impurities would not go in. The urine was then passed through the glass wool on the plastic case and diluted with 2000 ml of urine. Diluted urine was collected in a 50 ml conical tube and stored at -20 ° C before analysis. Moisture, crude protein, crude fat and crude ash were analyzed by AOAC (1995) method for chemical composition analysis of feed, feces and urine.

Morphology of small intestine

Three piglets of average weight were selected for each treatment and slaughtered at the 2nd and 5th weeks to obtain small intestines. For the histological examination after slaughter, 10 cm of the proximal duodenum, jejunum, ileum of the small intestine were collected. After removal of the digest, samples from the middle of each duodenum, jejunum, and ileum tissue were cut 6-8 cm, cleaned and stored in neutral-buffered formalin solution until further morphological analysis. The samples were then cut into two parts at each segment for cross and length section of intestine surface and then processed by the standard by the standard paraffin method. Sections (2~3cm) were stained with haematoxylin and eosin, and examined under a light microscope. It was used a Leica DM500 microscope with Leica DFC425 coupled to the computer program (Leica Application suite software) for image analysis. The villus height was measured as the distance between the crypt mouth and the tip of villi. The crypt depth was measured as the distance between the basement membrane and the mouth the crypt. The villi and crypts were expressed in

um (micrometer) units. In case of tissue photographs, the 2nd week small intestine was photographed 100 times and the 5th week small intestine was photographed 40 times.

Incidence of Diarrhea

Observation of diarrhea incidence was conducted every 8:00 am for 35 days. Data was recorded by each pen for 2 phases (phase 1 and phase 2). Score of diarrhea incidence was given into 4 numbers according to the condition of feces and diarrhea. (0 = normal feces; 1 = moist feces; 2 = mild diarrhea; 3 = severe diarrhea and watery diarrhea; Yen et al., 2015). Slightly wet feces on the rump area have been designated as contaminated piglets. After recording data, evidence of watery diarrhea was cleaned away every time to separate from next day.

Statistical and chemical analysis

Experimental diet and excreta were analyzed for contents of dry matter, ash, CP, and nitrogen by using the Kjeldahl procedure with Kjeltex (Kjeltex™ 2200, Foss Tecator, Sweden). Experimental data were analyzed as a randomized complete block (RCB) design using the General Linear Model (GLM) procedure of SAS. Each pen was used as the experimental unit (RCBD) in the feeding trial and individual pig was used as the experimental unit (CRD) in blood profiles and nutrient digestibility. The statistical model included two main effects, MCT levels and with or without organic acid. A significant difference for treatment effects was reported at $P < 0.05$ and a high significant difference was considered as $P < 0.01$.

Results and Discussion

Growth performance

The effects of medium chain triglyceride with organic acid on growth performance were presented in Table 3. During phase 1 (0 to 2 week), there was significant difference in ADG and G:F ratio by MCT effect (MCT, $P=0.04$, $P=0.01$). Also, an interaction effect (MCT x OA, $P=0.04$) and MCT effect (MCT, $P=0.01$) were observed in ADG during phase 2 (3 to 5 week). The interaction effect was resulted from the difference in the contents of MCT when 0.1% organic acid was added. Supplementing 0.1% MCT showed higher ADG than 0.2% MCT when 0.1% organic acid was added, while no difference was observed in ADG when no organic acid was added. Furthermore, ADG was affected by addition of MCT during whole experimental period (MCT, $P=0.01$). These results of increasing ADG and G:F ratio for weaning pigs were similar with the results of previous studies on MCT.

Most previous studies have suggested that the inclusion of MCT on the growth performance of piglets had a positive effect during phase 1 (Rodas and Maxwell, 1992; Hong et al., 2012; Loh et al., 2013; Han et al., 2011). Li et al. (2015) represented that all treatments containing MCT had higher ADG and G:F ratio compared to the control during phase 1. Similarly, Dierick et al. (2002) represented the two treatments containing MCT showed the highest ADG and statistically significant difference during phase 1.

The effects of MCT on ADG and feed efficiency improvement were due to the specific physiological and biological characteristics of MCT

compared with LCT (Li et al., 2005; Takeuchi et al., 2006). Furthermore, unlike LCT, MCT did not require carnitine because it passed through the dual mitochondrial membrane very rapidly (Sidossis et al., 1996). Therefore, MCT had an advantage that it could be used immediately as an energy source and give positive results when it was added in weaning pig diets to meet their energy need (Odle, 1997). Another reason was that there was a previous study that controlled the antibacterial response of pigs when adding MCT in feed. Dierick et al. (2002) reported that there were strong in vitro and in vivo antibacterial effects of MCFA in the pig proximal small intestine without growth-promoting antibiotics. In other words, MCT could reduce the number of bacteria which eventually led to more energy use for animal growth. Several effects of these complex MCT would contribute to better absorption capacity of the intestinal tract and lead to a better growth performance (Loh et al., 2013).

On the other hand, there was a significant difference in ADG in the interaction between MCT and organic acid during phase 2 in the experiment with supplementation of MCT and organic acid (MCT \times OA, $P=0.04$). In the previous study conducted by Kuang et al. (2015), feed intake was highly increased by supplementation of MCT with organic acid in the 21-day-old weaning pigs during 1 week and 2 weeks. Also, body weight, ADG, G:F ratio were increased by MCT with organic acid factor during phase 1 as well. The reason for good growth performance in the treatment with MCT with organic acid additions was due to the effects of MCT mentioned above as well as the effect of increasing the feed intake and growth rate of formic acid in organic acid (Lallès et al., 2009; Partanen, 2001).

Consequently, the supplementation of 0.1% MCT significantly improved ADG and feed efficiency. Furthermore, supplementing 0.1% MCT with 0.1% organic acid showed the greatest results in growth performance among all the treatments.

Blood profiles

The effects of medium chain triglyceride with organic acid on blood profiles were presented in Table 4. As a result of the analysis, there was no statistically significant difference in blood profiles during the whole experimental period.

Cortisol was a common measurement indicator of stress research (Cook et al., 1996). It was measured because Han et al (2010) reported that MCT could reduce the level of cortisol which was lower than even antibiotics. However, no statistically significant difference was observed in the current experiment.

IgG, TNF- α , IL-6, IL-1 β and IL-10 were the indicator of immunological responses. IgG was generally considered to be the most common type of antibody in blood circulation and plays an important role in controlling bacterial infections in the body (Haye and Karenegay, 1979; Hankins et al., 1994). TNF- α , IL-6 and IL-1 β exerted excessive inflammatory responses including the expression of inflammatory cytokine (Papada et al., 2013). IL-10 was an important immuno modulatory cytokine produced by many cell populations (Asadullah et al., 2003).

As a result, there was no significant difference in all the measurements of immune system. However, according to a previous study by Kuang et al. (2015), IL-1 β and IL-10 still had no significant difference

but IgG and TNF- α showed significant differences. The IgG values of treatment including MCT with OA improved significantly. They said that the immunity of weaned pigs was improved by MCT with organic acid consumption. TNF- α was significantly lower in the MCT with organic acid supplemented treatment. The reduction in feed intake and growth found in immunologically challenged or sick piglets were considered to be the result of increased activity of inflammatory cytokine such as TNF- α (Kelley et al., 1994). Most studies on piglets indicated that immunological problems could induce cytokine synthesis and secretion leading to protein catabolism (Webel et al., 1997). Therefore, the decrease in TNF- α secretion could be explained by the immunological approach to increasing feed intake and growth of piglets fed with MCT and organic acid supplementation (Kuang et al., 2015).

On the other hand, the results of this current experiment were not the same as those of Kuang et al., (2015) which was probably due to the large difference in MCT contents. The amount of MCT supplemented was 1 mg/kg or 2 mg/kg in the present study but 130 mg/kg was supplemented in the Kuang et al. (2015) study which included MCT at least 65 to 13 times more.

In conclusion, this experiment did not show significant differences in all the measurement of blood profiles. The increase of IgG and the increase of regulatory cytokine expression and the decrease of inflammatory cytokine expression which may be related to the proliferation of *Lactobacillus* and the reduction of *E.coli* with the supplementation of MCT and organic acid were expected but different results were showed because of different contents of MCT with OA.

Nutrient digestibility

The effects of medium chain triglyceride with organic acid on nutrient digestibility and nitrogen retention were presented in Table 5. As a result, no significant difference was found in nutrient digestibility and nitrogen retention among treatments.

However, unlike the results of this experiment, previous studies have shown that the supplementation of MCT improved nutrient digestibility. Han et al. (2010) reported that supplementing 0.1% MCT improved significantly CP (crude protein), Ca (calcium), P (phosphorus) and energy digestibility compared to the antibiotic treatment. Han et al. (2010) also reported that lipid affected nutrient digestibility by changing morphology of small intestine. In other words, MCT improved growth performance in the same experiment due to the result of high nutrient digestibility. In addition, Li et al. (2015) represented that the value of EE (ether extract) was statistically higher in the treatments added with MCT compared to the control. This was because MCT had the property of improving the mal-absorption of fat in the case of contracted absorptive surface or atrophied intestinal villi (Cancio and Menendez, 1964). Moreover, the treatment with 1.4% MCT resulted in positive DM (dry matter) digestibility. It was reported that this result was from an integrated improvement of digestibility of various nutrients.

In conclusion, there were many previous studies where supplementation of MCT resulted in improving nutrient digestibility, but no significant difference was found in this study with MCT and organic acid additions. The discrepancy may due to the differences in the composition of diet, the source, constituent, saturation degree and supplementation level

of dietary fat.

Intestinal morphology

The effects of medium chain triglyceride and organic acid on small intestinal morphology were presented in Table 6, figure 1 and 2. As a result, significant differences were observed in villus height in the duodenum (MCT, $P=0.04$) and villus height (MCT, $P=0.03$) in the ileum by MCT factor during phase 1 (0 to 2 week). In addition, duodenal villus height (OA, $P=0.01$) and villus height to crypt depth ratio (VH:CD; OA, $P=0.02$) were affected by supplementation of organic acid during phase 2 (3 to 5 week).

In summary, supplementing MCT increased villus height in the duodenum and ileum during phase 1 and supplementing OA increased duodenal villus height and villus height to crypt depth ratio during phase 2. These results of increasing villus height and VH:CD for weaning pigs were similar with the results of previous studies on MCT and organic acid.

Czernichow et al. (1996) noted that the supplementation of MCT could promote mucosal growth and enhance epithelial cell regeneration to create better-developed intestinal tracts. Loh et al. (2015) reported that supplementing MCT had greater duodenal and jejunal villus height compared with the control at 6 h, 6 and 8 days of age. The villus height in ileum at 8 days for MCT (1034.50 μm) was also significantly higher than the control (704.20 μm). Thus, MCT not only enhanced nutrient absorption and utilization, but also promoted the development of the intestines as a whole to improve growth performance. Consequently, the

increase in duodenal and ileal villus height by MCT factor during phase 1 in this current experiment was due to the fact that the supplementation of MCT improved villus height and strengthened the intestinal tracts as shown in the previous studies of Czernichow et al. (1996) and Loh et al. (2015).

Furthermore, the results of this current experiment that villus height was increased by OA factor during phase 2 were the same as those of Ferrara et al. (2015) and Hosseindoust et al. (2017). According to a previous study by Hosseindoust et al. (2017), supplementing organic acid numerically improved jejunal and ileal villus height at 35 days. Ferrara et al., (2015) also showed that villus height and crypt depth in jejunum were slightly higher in the treatments with organic acids and organic acid with MCT than in the control. The authors reported that the supplementation of organic acid and MCT may result in decreased apoptosis rates at the top of the villi. Therefore, the increase in villus height and VH:CD in duodenum by OA factor during phase 2 in this current experiment could be said to be the result of OA supplementation because it enhanced the villus height as shown in the previous studies of Hosseindoust et al. (2017) and Ferrara et al, (2015).

Therefore, the supplementation of MCT with organic acid like many previous studies influenced the morphological changes of the small intestine such as increasing villus height. In particular, MCT increased duodenal and ileal villus height significantly during phase 1 and OA increased significantly villus height and VH:CD in the duodenum during phase 2 in this current experiment. In other words, MCT and OA could increase villus height of small intestine which improved nutrient absorption

and utilization.

Incidence of diarrhea

The effects of medium chain triglyceride with organic acid on incidence of diarrhea were presented in Table 7. No significant difference was observed during the whole experimental period. However, diarrhea score in MCT treatments and MCT with OA treatments was lower than the control. This would be probably due to the effects of MCT and organic acid.

Yen et al. (2015) measured the diarrhea score and the number of diarrhea pigs with and without MCT supplementation (MCT 0% and 3%). As a result, the diarrhea score and the number of diarrhea pigs in the MCT supplemented treatment were lower compared to the control. In another previous study, researchers had noted that diarrhea occurred less in treatments with MCT (Tee et al., 2010). These results indicated that pathogenic bacteria including *Vibrio cholera*, *Salmonella typhimurium*, *Shigella sonnei*, *Hemophilus influenza* and enterotoxigenic *E. coli* were inactivated by MCFA or their monoglycerides (Petschow et al., 1998).

In addition, Tsioloyiannis et al. (2011) reported on the effect of organic acid on pig diarrhea by comparison of organic acid types. As a result, diarrhea was less in all treatments containing organic acid than in the control without organic acid. This was because piglets reduced intestinal pH by ingesting organic acids and inhibited the growth of bacteria, especially ETEC. In addition, previous studies have shown that the addition of organic acids in the diet reduced gastric pH and bacterial counts along the gastrointestinal tract (Cole et al., 1968; Thomlinson and

Lawrence, 1981).

Consequently, the difference was not statistically significant but the incidence of diarrhea was numerically reduced in all treatments with MCT or MCT with OA compared to the control.

Conclusion

When MCT was supplemented, it increased ADG in each phase and improved G:F ratio during phase 1. Furthermore, MCT significantly increased villus height in the duodenum and ileum during phase 1.

When organic acid was supplemented, it significantly increased duodenal villus height and villus height to crypt depth ratio during phase 2. However, no significant difference was observed in blood profiles, nutrient digestibility, and incidence of diarrhea.

When MCT with organic acid was supplemented, the better result was showed in ADG during phase 2. The interaction effect was resulted from the difference in the contents of MCT when 0.1% organic acid was added. Supplementing 0.1% MCT showed higher ADG than 0.2% MCT when 0.1% organic acid was added, while no difference was observed in ADG when no organic acid was added.

In conclusion, supplementing 0.1% MCT with 0.1% organic acid improved intestinal morphology, thus it had positive influence on growth performance in weaning pigs.

Table 1. Formula and chemical composition of weaning phase 1 (0-2 week)

Ingredients, %	Treatments ¹⁾				
	CON	LM	LMO	HM	HMO
Expanding corn	45.83	45.73	45.52	45.64	45.43
Soy bean meal	37.02	37.04	37.08	37.06	37.09
Soy-oil	1.84	1.81	1.88	1.79	1.86
Sweet whey powder	4.00	4.00	4.00	4.00	4.00
Lactose	8.00	8.00	8.00	8.00	8.00
L-Lysine-HCl, 78%	0.20	0.20	0.20	0.20	0.20
DL-met, 80%	0.05	0.04	0.04	0.04	0.04
L-threonine, 99%	0.04	0.04	0.03	0.03	0.04
MDCP	1.43	1.43	1.43	1.43	1.43
Limestone	1.06	1.06	1.06	1.06	1.06
Vit. Mix ²⁾	0.10	0.10	0.10	0.10	0.10
Min. Mix ³⁾	0.10	0.10	0.10	0.10	0.10
Salt	0.30	0.30	0.30	0.30	0.30
ZnO	0.05	0.05	0.05	0.05	0.05
MCT premix	0.00	0.10	0.10	0.20	0.20
Organic acid	0.00	0.00	0.10	0.00	0.10
Total	100.00	100.00	100.00	100.00	100.00
Chemical composition					
ME, kcal/kg ⁴⁾	3300.00	3300.01	3300.03	3300.01	3300.03
Moisture, % ⁵⁾	8.90	8.76	8.93	8.80	8.95
Crude protein, % ⁵⁾	18.50	19.24	20.42	20.17	20.18
Crude fat, % ⁵⁾	3.39	3.48	3.73	3.45	3.39
Crude ash, % ⁵⁾	7.18	5.97	5.88	6.06	5.87
Lysine, % ⁴⁾	1.35	1.35	1.35	1.35	1.35
Methionine, % ⁴⁾	0.35	0.35	0.35	0.35	0.35
Threonine, % ⁴⁾	0.86	0.86	0.86	0.86	0.86
Ca, % ⁴⁾	0.80	0.80	0.80	0.80	0.80
Total P, % ⁴⁾	0.65	0.65	0.65	0.65	0.65

¹⁾ CON : Corn-SBM based diet, LM : Corn-SBM based diet + MCT 0.1%, HM : Corn-SBM based diet + MCT 0.2%, LMO : Corn-SBM based diet + MCT 0.1% + organic acid 0.1%, HMO : Corn-SBM based diet + MCT 0.2% + Organic acid 0.1%.

²⁾ Provided the following quantities of vitamins per kg of complete diet : Vit A, 8,000IU; Vit D3, 1,800IU; Vit. E, 80IU; Vit. K3, 2mg; Ribo flavin, 7mg; Calcium pantothenic acid, 25mg; Niacin, 27mg; d-Biotin, 200ug; Vit.B12, 50ug.

³⁾ Provided the following quantities of minerals per kg of complete diet: Fe, 150 mg; Cu, 105 mg; Mn, 51 mg; I, 1 mg; Se, 0.3mg; Zn, 72 mg.

⁴⁾ Calculated value.

⁵⁾ Analyzed value.

Table 2. Formula and chemical composition of weaning phase 2 (3-5 week)

Ingredients, %	Treatments ¹⁾				
	CON	LM	LMO	HM	HMO
Expanding corn	57.11	57.02	56.82	56.93	56.73
Soy bean meal	32.29	32.31	32.33	32.32	32.35
Soy-oil	1.78	1.75	1.83	1.73	1.80
Sweet whey powder	2.00	2.00	2.00	2.00	2.00
Lactose	4.00	4.00	4.00	4.00	4.00
L-Lysine-HCl, 78%	0.11	0.11	0.11	0.11	0.11
DL-met, 80%	0.01	0.01	0.01	0.01	0.01
L-threonine, 99%	0.00	0.00	0.00	0.00	0.00
MDCP	1.22	1.22	1.22	1.22	1.22
Limestone	0.95	0.95	0.95	0.95	0.95
Vit. Mix ²⁾	0.10	0.10	0.10	0.10	0.10
Min. Mix ³⁾	0.10	0.10	0.10	0.10	0.10
Salt	0.30	0.30	0.30	0.30	0.30
ZnO	0.03	0.03	0.03	0.03	0.03
MCT premix	0.00	0.10	0.10	0.20	0.20
Organic acid	0.00	0.00	0.10	0.00	0.10
Total	100.00	100.00	100.00	100.00	100.00
Chemical composition					
ME, kcal/kg ⁴⁾	3300.01	3300.00	3300.01	3300.04	3300.02
Moisture, % ⁵⁾	9.02	9.24	9.33	9.22	8.78
Crude protein, % ⁵⁾	18.92	18.78	17.37	18.59	19.20
Crude fat, % ⁵⁾	3.97	3.78	3.53	3.29	3.61
Crude ash, % ⁵⁾	4.91	5.27	5.32	5.17	5.40
Lysine, % ⁴⁾	1.15	1.15	1.15	1.15	1.15
Methionine, % ⁴⁾	0.30	0.30	0.30	0.30	0.30
Threonine, % ⁴⁾	0.74	0.74	0.74	0.74	0.74
Ca, % ⁴⁾	0.70	0.70	0.70	0.70	0.70
Total P, % ⁴⁾	0.60	0.60	0.60	0.60	0.60

¹⁾ CON : Corn-SBM based diet, LM : Corn-SBM based diet + MCT 0.1%, HM : Corn-SBM based diet + MCT 0.2%, LMO : Corn-SBM based diet + MCT 0.1% + organic acid 0.1%, HMO : Corn-SBM based diet + MCT 0.2% + Organic acid 0.1%.

²⁾ Provided the following quantities of vitamins per kg of complete diet : Vit A, 8,000IU; Vit D3, 1,800IU; Vit. E, 80IU; Vit. K3, 2mg; Ribo flavin, 7mg; Calcium pantothenic acid, 25mg; Niacin, 27mg; d-Biotin, 200ug; Vit.B12, 50ug.

³⁾ Provided the following quantities of minerals per kg of complete diet: Fe, 150 mg; Cu, 105 mg; Mn, 51 mg; I, 1 mg; Se, 0.3mg; Zn, 72 mg.

⁴⁾ Calculated value.

⁵⁾ Analyzed value.

Table 3. Effects of medium chain triglyceride and organic acid on growth performance in weaning pigs¹⁾

Criteria	CON ²⁾	LM	LMO	HM	HMO	SEM ³⁾	P-value		
	MCT 0% OA 0%	MCT 0.1%		MCT 0.2%			MCT	OA	MCT•OA
		OA 0%	OA 0.1%	OA 0%	OA 0.1%				
Body weight, kg									
Initial	7.99	8.00	8.00	8.00	7.99	0.165	–	–	–
2 week	9.60	10.07	10.46	9.58	9.47	0.265	0.30	0.84	0.72
5 week	18.12 ^{bc}	19.52 ^{ab}	20.10 ^a	18.89 ^{abc}	17.75 ^c	0.382	0.09	0.73	0.31
ADG, g									
0–2 week	115	148	176	113	106	10.8	0.04	0.66	0.47
3–5 week	406 ^{bc}	450 ^{ab}	459 ^a	444 ^{ab}	394 ^c	8.6	0.01	0.13	0.04
0–5 week	289 ^{bc}	329 ^{ab}	346 ^a	311 ^{abc}	279 ^c	8.5	0.01	0.56	0.10
ADFI, g									
0–2 week	231	239	258	228	211	9.9	0.27	0.95	0.49
3–5 week	685	737	751	693	657	18.7	0.12	0.79	0.54
0–5 week	444	476	487	448	423	12.3	0.13	0.81	0.52
G:F ratio									
0–2 week	0.496	0.622	0.681	0.494	0.503	0.0286	0.01	0.52	0.74
3–5 week	0.592	0.610	0.611	0.640	0.601	0.0114	0.69	0.47	0.39
0–5 week	0.652	0.692	0.710	0.695	0.659	0.0107	0.38	0.68	0.24

¹⁾ A total 120 weaning pigs was fed from average initial body 8.00±0.694 kg and average final body weight was 18.87±2.160 kg.

²⁾ CON (Corn-SBM based diet), LM (Corn-SBM based diet + MCT 0.1%), LMO (Corn-SBM based diet + MCT 0.1% + Organic acid 0.1%), HM (Corn-SBM based diet + MCT 0.2%), HMO (Corn-SBM based diet + MCT 0.2% + Organic acid 0.1%).

³⁾ Standard error of the mean.

^{abc} Means in a same row with different superscript letters significantly differ (P<0.05).

Table 4. Effects of medium chain triglyceride and organic acid on blood profiles in weaning pigs

Criteria	CON	LM	LMO	HM	HMO	SEM ³⁾	P-value		
	MCT 0%	MCT 0.1%		MCT 0.2%			MCT	OA	MCT+OA
	OA 0%	OA 0%	OA 0.1%	OA 0%	OA 0.1%				
Cortisol, ng/ml									
Initial	-----	44.56		-----		—	—	—	—
2 week	5.48	16.24	4.68	25.36	10.14	3.551	0.29	0.06	0.78
5 week	24.33	14.68	10.58	20.93	18.28	2.887	0.27	0.58	0.90
IgG, g/ml									
Initial	-----	9.00		-----		—	—	—	—
2 week	10.43	11.73	10.35	11.01	10.68	0.492	0.85	0.43	0.63
5 week	15.67	19.04	14.14	15.15	17.39	1.113	0.88	0.56	0.13
TNF- α , pg/ml									
Initial	-----	127.89		-----		—	—	—	—
2 week	147.85	137.85	137.78	117.01	124.94	5.114	0.11	0.70	0.70
5 week	134.94	118.18	130.34	106.13	110.33	5.792	0.19	0.49	0.73
IL-6, pg/ml									
Initial	-----	317.56		-----		—	—	—	—
2 week	320.49	352.14	329.99	407.52	310.32	51.535	0.88	0.62	0.75
5 week	311.86	281.42	289.32	293.08	288.34	34.498	0.94	0.98	0.93
IL-10, pg/ml									
Initial	-----	7.94		-----		—	—	—	—
2 week	7.33	6.48	9.95	7.62	6.17	0.661	0.30	0.42	0.06
5 week	6.72	7.46	9.37	7.58	8.77	0.879	0.90	0.42	0.84
IL-1 β , pg/ml									
Initial	-----	23.61		-----		—	—	—	—
2 week	18.29	17.56	27.47	28.59	24.54	2.928	0.52	0.64	0.28
5 week	23.28	12.94	19.49	26.99	19.81	3.898	0.41	0.97	0.43

¹⁾ Least squares means of 5 observations per treatment.

³⁾ Standard error of the mean.

Table 5. Effects of medium chain triglyceride and organic acid on nutrient digestibility in weaning pigs.

Criteria	CON	LM	LMO	HM	HMO	SEM ²⁾	P-value		
	MCT 0%	MCT 0.1%		MCT 0.2%			MCT	OA	MCT•OA
	OA 0%	OA 0%	OA 0.1%	OA 0%	OA 0.1%				
Nutrient digestibility, %									
Dry matter	96.20	96.82	95.78	96.29	95.68	0.417	0.74	0.40	0.82
Crude protein	95.70	96.39	94.57	95.91	95.06	0.538	0.99	0.25	0.66
Crude ash	85.18	86.66	83.66	85.00	83.33	1.597	0.78	0.53	0.85
Crude fat	90.32	92.00	89.00	90.00	89.67	1.071	0.78	0.50	0.59
N-retention, g/d									
N-intake	70.51	69.82	64.51	69.12	71.74	2.557	—	—	—
N-feces	3.03	2.52	3.78	2.83	3.54	0.362	0.96	0.23	0.72
N-urine	2.14	1.76	2.63	1.96	2.54	0.258	0.92	0.21	0.79
N-retention ³⁾	65.34	65.54	63.25	64.34	65.65	0.633	0.65	0.72	0.20
N-retention ⁴⁾ , %	92.67	93.87	90.78	93.08	91.52	0.873	0.98	0.24	0.69

¹⁾ A total of 15 barrow and initial body weight 12.48 ± 0.37 kg

²⁾ Standard error of the mean.

³⁾ N retention = N intake (g) - Fecal N (g) - Urinary N (g)

⁴⁾ N retention (%) = N retention / N intake × 100

Table 6. Effects of medium chain triglyceride and organic acid on small intestine morphology in weaning pigs.

Criteria	CON	LM	LMO	HM	HMO	SEM ²⁾	P-value		
	MCT 0%	MCT 0.1%		MCT 0.2%			MCT	OA	MCT*OA
	OA 0%	OA 0%	OA 0.1%	OA 0%	OA 0.1%				
Weaning phase 1 (2week), μm									
Duodenum									
Villus height	312.67	340.26	341.08	291.83	251.87	14.687	0.04	0.51	0.49
Crypt depth	311.29	329.86	350.44	322.61	363.41	13.627	0.92	0.33	0.74
VH : CD ³⁾	1.00	1.03	0.97	0.90	0.69	0.062	0.13	0.29	0.47
Jejunum									
Villus height	276.31	314.60	329.08	280.93	311.27	12.723	0.41	0.47	0.79
Crypt depth	235.07	236.29	245.73	237.33	274.68	14.016	0.64	0.47	0.66
VH : CD	1.17	1.33	1.33	1.18	1.13	0.085	0.36	0.97	0.99
Ileum									
Villus height	231.82	272.76	292.61	234.63	253.37	10.877	0.03	0.24	0.97
Crypt depth	231.23	245.27	261.61	223.23	257.69	13.962	0.61	0.34	0.72
VH : CD	1.00	1.11	1.11	1.05	0.98	0.082	0.42	0.63	0.74
Weaning phase 2 (5week), μm									
Duodenum									
Villus height	273.63	243.07	313.97	272.59	295.06	12.321	0.72	0.01	0.13
Crypt depth	386.08	397.83	327.80	417.09	376.71	22.609	0.49	0.27	0.76
VH : CD	0.70	0.54	1.03	0.68	0.78	0.071	0.43	0.04	0.17
Jejunum									
Villus height	229.19	234.02	281.25	242.76	277.82	11.252	0.91	0.14	0.81
Crypt depth	291.93	298.49	250.48	242.08	266.93	11.035	0.36	0.59	0.11
VH : CD	0.78	0.78	1.12	1.00	1.04	0.056	0.45	0.10	0.24
Ileum									
Villus height	198.17	255.16	262.81	255.28	255.85	21.163	0.92	0.91	0.92
Crypt depth	215.74	242.83	246.05	242.16	230.89	15.841	0.83	0.91	0.84
VH : CD	0.91	1.05	1.06	1.05	1.10	0.092	0.94	0.82	0.77

¹⁾ Least squares means for three pigs per treatment.

²⁾ Standard error of the mean.

³⁾ VH : CD = villus height to crypt depth ratio.

	CON	LM	LMO	HM	HMO
	MCT 0%	MCT 0.1%		MCT 0.2%	
	OA 0%	OA 0%	OA 0.1%	OA 0%	OA 0.1%
Duodenum					
Jejunum					
Ileum					

Figure 1. Eosin (HE)-stained sections at duodenum, jejunum, and ileum of small intestine morphology in weaning pigs showing villous height and crypt depth (2 weeks after weaning)

	CON	LM	LMO	HM	HMO
	MCT 0%	MCT 0.1%		MCT 0.2%	
	OA 0%	OA 0%	OA 0.1%	OA 0%	OA 0.1%
Duodenum					
Jejunum					
Ileum					

Figure 2. Eosin (HE)-stained sections at duodenum, jejunum, and ileum of small intestine morphology in weaning pigs showing villous height and crypt depth (5 weeks after weaning)

Table 7. Effects of medium chain triglyceride and organic acid on incidence of diarrhea in weaning pigs

Criteria	CON	LM	LMO	HM	HMO	SEM ³⁾	P-value		
	MCT 0%	MCT 0.1%		MCT 0.2%			MCT	OA	MCT*OA
	OA 0%	OA 0%	OA 0.1%	OA 0%	OA 0.1%				
Diarrhea score ³⁾									
0-2 week	1.66	1.20	1.15	1.25	1.41	0.075	0.37	0.76	0.55
3-5 week	1.16	0.75	0.75	0.83	0.91	0.054	0.31	0.73	0.73
0-5 week	1.41	0.98	0.95	1.04	1.16	0.050	0.22	0.65	0.50

¹⁾ Least squares means for eight pigs per treatment.

²⁾ Standard error of the mean.

³⁾ Diarrhea score : 0 = normal feces, 1 = moist feces, 2 = mild diarrhea, 3 = severe diarrhea and watery diarrhea.

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V. Summary in Korean

본 연구는 자돈 사료 내에서 성장 촉진형 항생제 사용이 금지됨에 따라 성장 성적을 향상시키고 강한 항균 효과를 갖으며 영양소 소화율을 높인다는 공통점이 있는 MCT와 유기산제의 첨가에 따른 자돈에 있어 효과를 평가해보기 위해 이번 실험이 수행 되었다. 사양실험을 통해 성장성적, 혈액분석, 영양소 소화율, 소장의 형태학적 변화, 설사 빈도를 측정하였다. 사양실험을 위해 평균체중 8.00 ± 0.870 kg의 삼원교잡종 ([Yorkshire \times Landrace] \times Duroc) 이유자돈 120두를 공시하여 총 5주 동안 (자돈 전기 2주와 자돈 후기 3주) 실험을 수행 하였다. 실험돈들은 체중과 성별에 따라 5처리 3반복, 펜 당 8마리씩 난괴법 (RCBD; randomized complete block design)으로 배치하여 수행되었다. 실험은 2×2 factorial arrangement + control 로 설계 되었으며, 실험 처리구는 다음과 같다. 요인 1은 사료 내 MCT 함량으로 0.1% (1,000 ppm)와 0.2% (2,000 ppm)로 구성되었으며, 요인 2는 사료 내 유기산제 첨가 유무로 0%와 0.1%로 구성되어 있다. 이에 따라 1) CON : NRC (1998)의 영양소 요구량을 충족하는 옥수수-대두박 위주의 기초 사료, 2) LM : 기초사료 + MCT 0.1 % 첨가, 3) LMO : 기초사료 + MCT 0.1 % + OA 0.1% 첨가, 4) HM : 기초사료 + MCT 0.2 % 첨가, 5) HMO : 기초사료 + MCT 0.2 % + OA 0.1%로 구성 되어 있다.

사양실험 결과, 성장성적에 있어 자돈 전기 때 0.1% MCT 첨가가 유의적으로 일당증체량 (MCT, $P=0.04$)과 사료효율 (MCT, $P=0.01$)을 향상시켰다. 자돈 후기 때에도 0.1% MCT 첨가 그리고

MCT와 유기산제의 교호효과에 의해 일당증체량 (MCT, $P=0.01$; MCT \times OA, $P=0.04$)에서 유의적인 차이가 발생했으며 특히 0.1% MCT와 0.1% 유기산제를 첨가한 처리구가 다른 처리구들에 비해 수치적으로 체중, 일당증체량, 일일사료섭취량, 사료효율에서 가장 좋은 성장성적을 보였다. 소장의 형태학적 변화에 있어서는 자돈 전기 때는 MCT에 의해 십이지장 (MCT, $P=0.04$)과 회장의 용모 길이 (MCT, $P=0.03$)에서 유의적인 차이가 나타났으며 자돈 후기 때는 유기산제에 의해 십이지장의 용모 길이와 용모 길이:용와 깊이 비율에서 유의적인 차이가 나타났다 (OA, $P=0.01$, $P=0.04$). 특히 0.1% MCT와 0.1% 유기산제가 첨가된 사료를 급여 받은 자돈들은 다른 처리구의 자돈들보다 더 긴 용모 길이를 보였다. 실험 전 기간 동안 혈액성상, 영양소 소화율, 설사 빈도에 있어서는 통계적인 유의적 차이가 나타나지 않았다.

결론적으로 0.1% MCT와 0.1% 유기산의 첨가는 장 형태를 향상시켜 이유자돈의 성장성적에 있어 긍정적인 영향을 미쳤다.